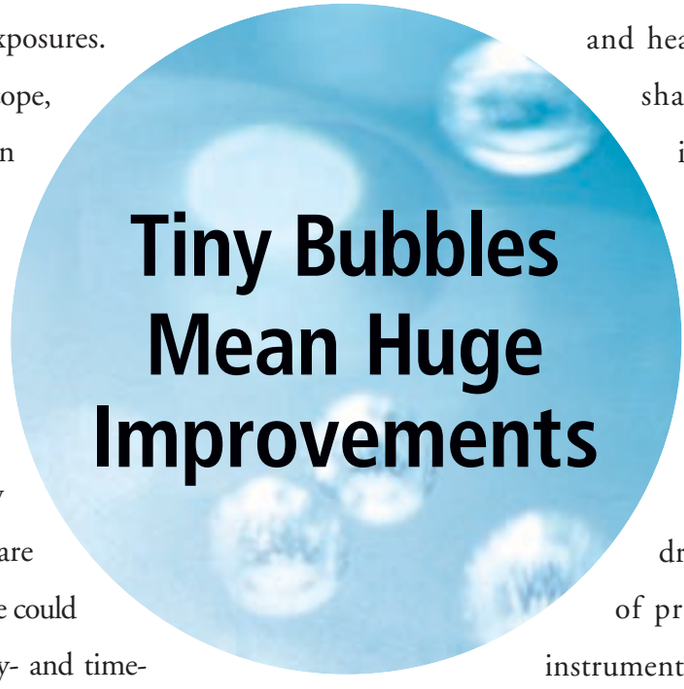
A Cleaner Doctor's Kit

Tiny Bubbles Mean Huge Improvements



Medical and dental instruments are composed of a variety of materials, some that can withstand repeated applications of heat and chemical treatment, and others that will fail, quickly and completely, upon such exposures. Something like an endoscope, which can cost more than $30,000, is a little pricey to treat as a disposable commodity, and the environmental costs of current cleaning techniques can be high. Now two Georgia researchers are developing what they believe could be a safer and more energy- and time-efficient alternative to traditional technologies without the use of harmful chemicals or damaging heat.

Doctors currently use a number of sterilization technologies, each of which carries its own pluses and minuses. The steam autoclave is one of the most popular technologies because it uses no special chemicals, has a relatively short cycle time compared to other methods, and poses no environmental hazards. The downside is the combination of moisture and heat, which can reduce the sharpness and longevity of instruments. And some studies have shown evidence of biofilms forming in steam autoclaves and emitting heat-stable toxins.

A second technology, dry heat, has the advantage of producing completely dry instruments, with a correspondingly lower impact on sharpness. However, the requisite higher temperatures demand more energy to reach and sustain, and, combined with longer cycle times, create greater and more rapid wear on the instruments being sterilized.

Brand X Pictures, Photodisc, Matt Ray/EHP

A third technology is unsaturated chemical vapor sterilization, which relies on a mixture of chemicals such as formaldehyde and alcohol. This technology has the advantage of shorter cycles and minimal rusting and dulling, but has serious environmental downsides because of chemical use and disposal issues and potential health effects. Formaldehyde, for example, is classified as a potential human carcinogen and can cause health effects ranging from irritation of the eyes, nose, and throat to severe asthmatic reactions, chest pain, and shortness of breath. Thus, this technique has fallen out of favor.

Bring On the Bubbles

Stephen Carter, an Atlanta-area dentist, and Ken Cunefare, an engineering professor at the Georgia Institute of Technology, are currently developing what could be an excellent alternative to traditional cleaning processes. The new method relies on cavitation, where acoustic energy causes the formation in liquid of bubbles, which release energy as they collapse. This phenomenon was first studied because of its impact on submarines—cavitation damages the propellers of submarines, and the collapse of the bubbles creates a noise that is detectable by enemies.

The energy released by the collapsing bubbles increases dramatically with an increase in hydrostatic pressure to a certain point (typically about twice normal atmospheric pressure), and then the energy begins to fall off dramatically, a property known as the "anomalous depth effect." Because the bubbles collapse with far greater intensity at optimal elevated pressures, Carter and Cunefare call the resulting phenomenon "enhanced transient cavitation."

Carter explains that debris in the liquid or on the container walls will act as a nucleation site for bubble formation. "A single virus or bacterium probably won't have the mass to trigger this formation," he says. "But bubble formation will occur in the [liquid] nonetheless. It just takes place a little more readily in the presence of physical matter."

Further, says Carter, the bubbles that form are attracted to microbes. If a bubble forms within 0.1 millimeter of a microbe, an asymmetrical expansion is triggered—the bubble shoots out a jet that fractures the cell wall.

An idea bubbles to the surface. Ken Cunefare holds a test chamber used to test the ability of ultrasonic cavitation to kill bacteria.

Carter has been experimenting with cavitation for several years. In 1994, he received a patent for the use of "explosive decompression" as a means of sterilizing medical devices. "The idea was that you'd put contaminated instruments into an enclosed chamber, then increase pressure in the chamber while also introducing an antimicrobial solution," Carter explains. "That increase in pressure would allow the solution to be forced through the microbial walls, and then you'd suddenly decompress the chamber, thus rupturing the cell walls and killing the microbes."

The technology worked pretty well, except with spores, the most durable forms of bacteria. The spore wall, which is designed to help the microbe survive long periods in a hostile environment, wouldn't fracture under these circumstances.

The next step, Carter reasoned, was to see what would happen if he combined pressure, an antimicrobial solution, and sound. "Even at ambient pressure, ultrasonic cavitation is far more effective at killing germs than the solution on its own," says Carter. He hoped that elevated pressure would amplify the germ-killing properties. Ultrasound, which he discovered worked best at around 30,000 cycles per second, produced a series of pressure waves that hammered against the tough spore wall. He received a patent on this method in 1997. But the kill levels still were not satisfactory.

That's when Carter teamed up with Cunefare. Together, they worked to find an optimal combination of ultrasonic energy, pressure, and antimicrobial concentration using a small chamber devised to hold 80 cubic centimeters of solution and spore forms of nonpathogenic *Bacillus stearothermophilus* and *Bacillus subtilis*.

According to Carter, their initial tests revealed an undetectable kill level in the solution at ambient pressure and without ultrasound. Adding ultrasound alone resulted in only a 3% bacterial kill rate over a 10-minute test period. But when the container was pressurized to 30 pounds per square inch, the kill rate rose to 90% within 1 minute. (Beyond 1 minute, Carter notes, heat buildup could have skewed test results.) Experimentation eventually determined that a 68% solution of isopropyl alcohol was most effective. These results were

Gary Meek/Georgia Institute of Technology

presented in December 2002 at the First Pan-American/Iberian Meeting on Acoustics.

"This was encouraging," Carter says, "because the device we used to generate the ultrasound had a ten percent 'duty cycle' [on for one-tenth of a second, off for nine-tenths of a second]. What we're looking for now is a way to get a one hundred percent duty cycle, which could give us sterilization in four to five minutes [compared to up to two hours required by current methods]. Ultrasonic generators which can function at one hundred percent duty cycle are available."

Toil and Trouble?

Despite encouraging signs, Carter points out there are many issues yet to be addressed, and robust efficacy studies must be concluded before the Food and Drug Administration would approve a new sterilizer such as the Georgia team proposes.

"For one thing," Carter says, "we still need to run more tests to confirm this process won't damage things like seals or adhesives, and won't pit delicate instruments with repeated usage. And we need to develop a mechanism to pump in refrigerant for the next phase of testing, so the solution temperature won't exceed one hundred degrees Fahrenheit."

It is also possible, he says, that different alcohol concentrations will yield a more effective process, because increasing the volatility of the solution increases the rate of cavitation; tests in this area are still ongoing. The addition of volatile liquids to enhance cavitation is the subject of a second patent related to this process, which was recently granted to Carter and Cunefare.

Another aspect of cavitation to be considered is the momentary, yet intense, burst of heat generated as each bubble collapses. Temperatures can reach tens of thousands of degrees in a microsecond, temperatures certainly high enough to damage or destroy any medical equipment.

But Carter says other factors combine to lessen the impact of these heat bursts, including the fact that the bursts are very highly localized. "They won't heat up the solution if a cooling system is in place," he says. The team is currently working on an apparatus with such a cooling system.

Another unknown is the actual mechanism of microbial destruction. Despite researchers' best guesses, and although the end result is apparent, it's still unknown exactly how cavitation kills cells. "Increased pressure and disinfectant molecules are somehow enhanced by the cavitation process," said Donald Ahearn, professor emeritus of biology at Georgia State University, who performed the bioassays during the early studies, in the winter 2003 issue of Georgia Tech's *Research Horizons* magazine, "but the physiology of the death has yet to be determined."

Finally, some experts would like to see testing with prions, a type of protein that can become "misfolded" and contribute to brain diseases including variant Creutzfeld-Jakob disease and *kuru*, a fatal dementia among a tribe in New Guinea caused by eating the remains of diseased humans. Prion research is still quite new, but studies suggest that these misfolded proteins clump together, killing cells and, eventually, the organism, and that they are resistant to high temperatures.

William Keevil, director of the Environmental Healthcare Unit in the School of Biological Sciences at Southampton University, says, "My concern is that sonication cavitation may break up prion aggregates, but not destroy the robust prion protein, effectively amplifying the number of infectious units. It is essential, therefore, that someone determines whether the cavitation proposed can actually destroy the prion protein, and not just amplify larger numbers of smaller infectious aggregates."

According to Charles Palenik, chairman of the board of directors for the Organization for Safety & Asepsis Procedures, the kill figures observed to date still don't qualify as sterilization, "so that will either have to improve, or the system will have to be used as a precleaning system, which would certainly be of value in the sterilization process." (Although the literal definition of a sterile medical device is one that is free of viable microorganisms, there are different standards around the world for what constitutes sterility, and the definition may further vary depending on the intended use of the instrument.)

Obviously, says Palenik, it's too early to tell what the cost of such a cavitation system might be, but to make it attractive to private practitioners, it would need to be in the $1,500–3,000 range.

Rising to the Top

Despite the issues that remain to be ironed out, cavitation could hold great promise for the future of medical and dental instrument sterilization. Ultrasonic cleaning is already in use in many applications, from devices to clean jewelry to experimental drug delivery methods. Carter feels if this technology continues to prove out the way it has to date, it will open up a whole range of new, safer, and more energy-efficient applications, perhaps in areas such as wastewater treatment and low-temperature pasteurization of products such as milk and orange juice.

Lance Frazer

Suggested Reading

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